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Indian Standard

METHOD FOR TESTING FLAX FABRICS FOR RESISTANCE TO ATTACK BY MICRO-ORGANISMS

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NEW DELHI 110002

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Indian Standard

METHOD FOR TESTING FLAX FABRICS FOR RESISTANCE TO ATTACK BY MICRO-ORGANISMS

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Indian Standard

METHOD FOR TESTING FLAX FABRICS FOR RESISTANCE TO ATTACK BY MICRO-ORGANISMS

0. FOREWORD

0.1 This Indian Standard was adopted by the Indian Standards Institution on 8 December 1966, after the draft finalized by the Textile Chemistry Sectional Committee had been approved by the Textile Division Council.

0.2 The flax fabrics are mainly used for making hoses used by fire services for pumping water for extinguishing fire outbreaks. Like cotton, flax is also liable to deterioration by the action of micro-organisms. Since fire hoses are often stored, transported and used under conditions favourable for growth of micro-organisms, protective treatments are applied.

0.3 Studies on the pattern of microbial degradation of flax fabrics in soil have revealed the predominant activity of the two fungi, namely, *Pencillium* sp and *Memnoniella echinata.* These two fungi cause more than 90 percent loss in the breaking strength when used in ' pure culture ' test in 42 days. Whilst in soil enriched with these fungi, flax fabrics register 100 percent loss in breaking strength in 28 days.

0.4 Two quantitative methods, namely, ' soil burial ' method and ' mixed culture ' method, have been prescribed in this standard. The soil burial method is intended for testing rotproofness of the treatment, involving burial in composted soil enriched with the two specific fungi; while the mixed culture method determines the resistance of the flax fabric to fungal attack by infecting the fabric with *Penicillium* sp and *Memnoniella echinata.*

0.5 For evaluating the fabrics which had received treatment, an authentic sample of the basic untreated fabric is necessary.

0.6 The methods prescribed in this standard may also be used for evaluating preservatives or treatments designed to protect flax fabrics from damage due to micro-organisms.

0.7 Cultures of the organisms prescribed in this standard for this test may be obtained from the Director, Defence Research Laboratory (Materials), P. B. No. 320, Kanpur. To facilitate supply, the culture numbers as referred below may also be mentioned:

MICRO-ORGANISM	D.R.L. (M) CULTURE NO.
Memnoniella echinata	843
Penicillium funiculosm	844

0.8 In reporting the results of a test or analysis made in accordance with this standard, if the final value, observed or calculated, is to be rounded off, it shall be done in accordance with IS : 2-1960*.

1. SCOPE

1.1 This standard prescribes methods for evaluating flax fabrics for resistance to attack by micro-organisms.

2. SAMPLING

2.1 Lot — The quantity of flax fabrics of the same type and quality delivered to a buyer against a despatch note shall constitute a lot.

2.2 Unless otherwise agreed to between the buyer and the seller the number of pieces to be selected at random from a lot shall be as given below:

Lot Size	*Sample Size*
Up to 150	3
151 „ 300	4
301 „ 500	5
501 and above	7

2.3 The pieces thus selected shall constitute the **test sample**.

2.4 From the basic fabric (*see* **0.5**), take a sufficiently long piece. The piece thus taken shall constitute the **control sample**.

3. SOIL BURIAL METHOD

3.1 Test Soil — For the purpose of this method, prepare test soil in the manner prescribed in Appendix A.

3.2 Spore Suspension

3.2.1 Prepare the spore suspension of the following organisms in the manner prescribed in Appendix B:

a) *Memnoniella echinata*, and

b) *Penicillium* sp.

3.3 Procedure

3.3.1 From each piece in the test sample, cut out, at random, at least 3 test specimens so that the total number of test specimens obtained is at

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*Rules for rounding off numerical values (*revised*).

least 15. Each test specimen shall be 325 × 60 mm in size with the longer side parallel to warp threads. Mark these test specimens suitably for identification. The test specimens thus obtained shall be divided into 3 equal groups each containing at least one test specimen from each piece.

3.3.2 Take the test specimens belonging to the first group and determine the breaking load of each test specimen by the method prescribed in **9.1** of IS:1969-1961*. Find the average of all the test values.

NOTE — The size of the test strip should be 200 × 50 mm.

3.3.3 Put the test soil in different 1-litre glass jars (17 cm in height and 6 cm in diameter) having metallic screw caps. Spray 5 ml of the mixture of two spore suspensions on the test soil (*see* Note). Bury one test specimen belonging to the second group in each jar in such a way that the test specimen takes up 'U' shape inside the soil with two free ends (each 5 cm) projecting above the soil surface. Incubate the specimens at 30° ± 2°C and 98 ± 2 percent relative humidity for 28 days. At the end of this period, remove the specimen, wash free from soil, dry them in air and determine the breaking load of each specimen by the method prescribed in **9.1** of IS : 1969-1961*. Find the average of all the test values.

NOTE — While preparing soil compost the mixture of suspension is added to the soil and moistened adequately to have specified moisture content.

3.3.4 Leach the test specimens belonging to the third group in the manner prescribed in Appendix C and test them in the manner prescribed in **3.3.3**.

3.3.5 From the control sample cut at random 10 test specimens each measuring 325 × 60 mm with their longer sides parallel to warp threads. Take five specimens and determine the breaking load of each individual specimen by the method prescribed in **9.1** of IS : 1969-1961*. Find the average of all the test values.

3.3.6 Test the remaining five specimens in the manner prescribed in **3.3.3**.

3.3.7 Calculate the loss in breaking load, percent, of the basic fabric, on soil burial, by the following formula:

$$L = \frac{100\ (a - b)}{a}$$

where

L = Loss in breaking load, percent, of the basic fabric on soil burial,

a = Average breaking load value obtained as in **3.3.5**, and

b = Average breaking load value obtained as in **3.3.6**.

NOTE — The value of L should be 90 percent or more. If this value is less than 90 percent, the test should be repeated.

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*Method for determination of breaking load and elongation at break of woven fabric (by constant-rate-of-traverse machine).

3.3.8 Calculate in the manner prescribed in **3.3.7**, the loss in breaking load, percent, of the fabric under test on *soil burial without prior leaching* but equating to:

'*a*', value obtained as in **3.3.2**, and
'*b*', value obtained as in **3.3.3**.

NOTE — The value of *L* should not be more than 10 percent. The fabric should be considered satisfactorily resistant to attack by micro-organisms if the value of *L* does not exceed 10 percent.

3.3.9 Calculate in the manner prescribed in **3.3.7** the loss in breaking load, percent, of the fabric under test on *soil burial after leaching* but equating to:

'*a*', value obtained as in **3.3.2**, and
'*b*', value obtained as in **3.3.4**.

NOTE — The value of *L* should not be more than 10 percent. The fabric should be considered satisfactorily resistant to attack by micro-organisms if the value of *L* does not exceed 10 percent.

3.4 Report the results of the test as under:

a) Loss in breaking load, percent of the basic fabric on soil burial (*see* **3.3.7**);
b) Loss in breaking load, percent of the fabric under test on soil burial without prior leaching (*see* **3.3.8**); and
c) Loss in breaking load, percent of the fabric under test on soil burial after leaching (*see* **3.3.9**).

4. MIXED CULTURE METHOD

4.1 Apparatus — For the purpose of this test, petri dishes each 15 cm in diameter and 4 cm in height shall be used.

4.2 Test Organisms — For the purpose of this test, cultures of the following organisms shall be used:

a) *Memnoniella echinata*, and
b) *Penicillium* sp.

4.3 Test Medium — prepared by mixing the following:

Ammonium nitrate	3·0 g
Potassium dihydrogen phosphate	0·25 g
Magnesium sulphate	0·25 g
Potassium chloride	0·25 g
Agar-agar	25 g

in one litre of distilled water (*see* IS : 1070-1960*) and sterilizing with

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*Specification for water, distilled quality (*revised*).

steam at a pressure of 1·1 kg per square centimetre for 20 minutes.

NOTE 1 — Only 'pure chemicals' shall be used for preparing medium. 'Pure chemicals' shall mean chemicals that do not contain impurities which affect the results of the test.

NOTE 2 — The pH of the medium should be 6·4.

4.4 Procedure

4.4.1 From each piece in the test sample, cut out at random at least 2 test specimens so that the total number of test specimens obtained is at least 10. Each test specimen shall be 325 × 60 mm in size with the longer side parallel to the warp threads. Mark these test specimens suitably for identification. The test specimens thus obtained shall be divided into 2 equal groups each containing at least one test specimen from each piece.

4.4.2 Prepare in the manner prescribed in Appendix B a mixed suspension of spores of the test organisms prescribed in **4.2**.

4.4.3 Take the test specimens belonging to the first group. Sterilize the specimens in the manner prescribed in Appendix D and with sterile instruments transfer them to different sterile Petri dishes each containing 60 to 70 ml of test medium. Roll up the two ends of the test specimens so that about 10 cm of the central portion is left exposed. Keep one test specimen in each dish. Inoculate the exposed portion of each specimen in steriled condition with 3 to 4 ml of the mixed spores suspension (*see* **4.4.2**). Incubate the specimens for 28 days at 30° ± 2°C and 90 ± 2 percent relative humidity. At the end of the period, remove the specimens and record the degree of fungal growth as per the rating given in Table 1. Wash the test specimens with cold water till they are free from agar and determine the breaking load of each by the method prescribed in **9.1** of IS: 1969-1961*. Find the average of all the values.

4.4.4 Leach the test specimens belonging to the second group in the manner prescribed in Appendix C and test them in the manner prescribed in **4.4.3**.

4.4.5 From the control sample, cut at random five control specimens and test them in the manner prescribed in **4.4.3**.

4.4.6 Calculate the loss in breaking load, percent of the basic fabric *on inoculation* by the following formula:

$$L = \frac{100\,(a - b)}{a}$$

---

*Method for determination of breaking load and elongation at break of woven fabric (by constant-rate-of-traverse machine).

where

L = Loss in breaking load, percent, of the basic fabric on inoculation;

a = Breaking load, value obtained as in **3.3.5**; and

b = Breaking load value obtained as in **4.4.5**.

NOTE — The value of L should be 60 percent or more. If this value is less than 60 percent, the test should be repeated.

TABLE 1 DEGREE OF FUNGAL GROWTH

(*Clause* 4.4.3)

SYMBOLIC RATING (1)	STATE OF GROWTH OF MICRO-ORGANISMS (2)	RATING IN TERMS OF RESISTANCE (3)
—	No growth	Good
±	Very slight growth (visible only under magnifying glass)	Very Fair
+	Slight growth (area of growth less than one-fourth of total)	Fair
++	Moderate growth (area of growth more than one-fourth but less than half)	Poor
+++	Heavy growth (area of growth more than half)	Very Poor

4.4.7 Calculate in the manner prescribed in **4.4.6** the loss in breaking load, percent, of the fabric under test, *on inoculation without prior leaching* but equating to:

'a', the value obtained as in **3.3.2**; and

'b', the value obtained as in **4.4.3**.

NOTE — The value of L should not be more than 10 percent. The fabric should be considered satisfactorily resistant to attack by micro-organisms if the value of L does not exceed 10 percent.

4.4.8 Calculate in the manner prescribed in **4.4.6** the loss in breaking load, percent, of the fabric under test, *on inoculation after leaching* but equating to:

'a', the value obtained as in **3.3.2**; and

'b', the value obtained as in **4.4.4**.

NOTE — The value of L should not be more than 10 percent. The fabric should be considered satisfactorily resistant to attack by micro-organisms if the value of L does not exceed 10 percent.

4.5 Report the results of the test as under:

a) Loss in breaking load, percent, of the basic fabric on inoculation (*see* **4.4.6**),

b) Loss in breaking load, percent, of the fabric under test on inoculation without prior leaching (*see* **4.4.7**), and

c) Loss in breaking load, percent of the fabric under test on inoculation after leaching (*see* **4.4.8**).

APPENDIX A

(*Clause* 3.1)

PREPARATION OF TEST SOIL

A-1. PROCEDURE

A-1.1 Mix thoroughly:

a) a layer of good, sandy loam soil, 13 cm thick;
b) a layer of fresh cow-dung manure 7·5 cm thick; and
c) a layer of finely chopped straw 15 cm thick.

Allow the mixture to ferment for 2 months turning it once a week, maintaining the moisture content at 22 to 25 percent on the total weight of the mixture. At the end of the period, sift the resulting compost through 3·35-mm IS Sieve (*see* IS : 460-1962*) and test it in the manner prescribed below.

A-1.1.1 Take a light-weight cotton fabric weighing 240 to 255 g per square metre. Draw six 325 × 60 mm pieces from it. Take three pieces and determine the breaking load of each individual piece by the method prescribed in **9.1** of IS : 1969-1961†. Find the average of all the values.

A-1.1.2 Bury the remaining three pieces in the compost (*see* **A-1.1**) for 5 days. At the end of the period, remove the pieces from the compost and determine the breaking load of each individual piece by the method prescribed in **9.1** of IS :1969-1961†. Find the average of all the values.

A-1.2 Calculate the loss in strength of the fabric after soil burial.

NOTE — The soil should be deemed to be suitable for test if the loss in strength of fabric so determined is 80 percent or more.

APPENDIX B

(*Clauses* 3.2.1 *and* 4.4.2)

PREPARATION OF SPORE SUSPENSION

B-0. GENERAL

B-0.1 If a mixed suspension of spores of more than one test organism is to be prepared, the suspensions of spores of each test organism shall be prepared separately and the suspensions then mixed in equal volumes.

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*Specification for test sieves (*revised*).

†Method for determination of breaking load, and elongation at break of woven fabric (by constant-rate-of-traverse machine).

B-1. PREPARATION OF CULTURE MEDIUM

B-1.1 Take sufficient quantity of potatoes, say, about 250 g, peel them off and cut them into slices. Weigh 200 g of the sliced potatoes, wash them in running tap water and place them in a beaker; add 500 ml of distilled water and keep the contents at boil for 30 minutes. Filter the hot extract through a thin layer of absorbent cotton previously wetted with distilled water. Preserve the filtrate.

B-1.2 Take 25 g of agar, cut it into small pieces and boil, in the minimum quantity of distilled water necessary till no solid agar is visible. Add 20 g of dextrose and mix it with the filtrate (*see* **B-1.1**). Make up the volume to 1 000 ml with hot distilled water. Adjust the *p*H of the culture medium to 6·4.

NOTE — To avoid solidification, the temperature of the culture medium should be kept above 45°C.

B-1.3 Put 5 ml of the culture medium into each of a number of 15 × 2 cm test-tubes and plug them with non-absorbent cotton-wool. In an autoclave, sterilize the culture medium with steam at a pressure of 1·1 kg/cm^2 for 20 minutes. At the end of the period, remove the test-tubes from the autoclave, place them in a slanting position and allow them to cool to harden the culture medium into ' slants '.

B-2. PREPARATION OF SUB-CULTURE

B-2.1 Inoculate the slants in the test-tubes with a well-sporulated culture of test organism (*see* **3.2.1**) by means of a sterile needle. Incubate the slants for 7 days at 30° ± 2°C.

NOTE — At the end of this period, a heavy growth of the freshly sporulating test organism should be visible within the tubes.

B-3. PREPARATION OF SPORE SUSPENSION

B-3.1 Add about 10 ml of sterile distilled water to freshly sporulating sub-culture of the test organism (*see* **B-2**) within the tubes. Bring the spores of the sub-culture into suspension by shaking or disturbing them gently with a sterile glass rod. Filter the suspension through a sterile muslin or similar fabric to remove particles of agar or fungal mycelium. Examine a drop of the spore suspension under a low power microscope, and see whether 50 or more spores appear in the field.

APPENDIX C

(*Clauses* 3.3.4 *and* 4.4.4)

METHOD OF LEACHING

C-1. PROCEDURE

C-1.1 Place the specimen inside a glass jar with internal diameter of 9 cm. Through a capillary, extending to within 0·6 cm of the bottom of the jar, allow water (with pH of not less than 6·0 and not more than 8·0 and temperature of 30° ± 2°C) to flow into it at the rate of 10 litres per hour. In this stream of water, leach the specimen for 24 hours.

APPENDIX D

(*Clause* 4.4.3)

STERILIZATION IN METHYL ALCOHOL VAPOUR

D-1. PROCEDURE

D-1.1 Pack the specimens loosely in a desiccator of suitable size containing methyl alcohol and keep them for 24 hours (or longer) at room temperature (about 27°C). In the meantime, make another desiccator ready by sterilizing it with methyl alcohol vapour for a period of 12 hours at room temperature. Transfer the specimens from the first desiccator to the second desiccator with a pair of sterile tongs. Connect the second desiccator with a water pump through a cotton-wool filter and exhaust the desiccator at room temperature for 6 hours to remove traces of methyl alcohol. Bring the pressure in the desiccator to the atmospheric pressure by slowly letting in air through the cotton-wool filter.

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